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毛茛科鸭跖花属的核形态研究

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Karyomorphology of the genus *Oxygraphis* Bunge (Ranunculaceae)

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Abstract Three species of the genus *Oxygraphis* Bunge (Ranunculaceae) were karyomorphologically studied. The interphase nuclei of complex chromocenter type, prophase chromosomes of interstitial type and basic chromosome number of $x = 8$ were common to all the three species examined, but the chromosomes of *O. delavayi* were markedly larger than those of *O. glacialis* and of *O. tenuifolia*. The results support the viewpoints that *Oxygraphis* is a natural member of the subtribe Ranunculinae Spach, tribe Ranunculeae DC. in the subfamily Ranunculoideae and that *O. delavayi* might be the primitive species of this genus.

Key words Chromosome; *Oxygraphis* Bunge

1 Introduction

Oxygraphis Bunge (Ranunculaceae) is a small genus of four species. The most widely distributed species, *O. glacialis* (Fischer ex DC.) Bunge, occurs in Asia and North America. *O. endlicheri* (Walp.) Bennet et Chandra (= *O. polypetala* (Royle) Hook. f. et Thoms.) is distributed in the Himalayas and western China, while *O. tenuifolia* W. E. Evans and *O. delavayi* Franch. are both endemic to the Hengduan Mountains in western China (Tamura, 1995; Wang, 1995).

Gross-morphologically *Oxygraphis* is closely related to *Ranunculus* L. (Wang, 1995). Both of them are put together with several other related genera in the subtribe Ranunculinae Spach, tribe Ranunculeae DC. in the subfamily Ranunculoideae (Tamura, 1995).

Karyomorphology of this small genus is quite little known. Johnson and Packer (1968) reported the chromosome number of $2n = 16$ for *O. glacialis*. Yang and Wu (1993) confirmed this chromosome number and reported the karyotype as $2n = 6m + 6sm + 4st$, with the eighth chromosome pair being submedian-centromeric. This result has drawn my attention and I consider it quite doubtful, as in the tribe Ranunculeae of the Ranunculaceae the eighth chromosome pair is usually subterminal-centromeric (Goepfert, 1974; Kurita, 1957). This paper is to re-examine the chromosomes of this species and other two species in this genus for a better understanding of the systematic position of the genus and the chromosomal differentiation among the species.

2 Materials and Methods

Living plants were collected from the field and were grown in Kunming Botanic Garden, Kunming Institute of Botany, the Chinese Academy of Sciences. Localities and sources of the three spe-

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cies studied are given in Table 1. The voucher specimens are deposited in the Herbarium of Institute of Botany, the Chinese Academy of Sciences (PE).

For the observation of the chromosomes, vigorously growing roots were harvested and then pretreated with 0.1% colchicine for 2.5 hours at room temperature. They were then fixed in Carnoy's Fixative (absolute ethanol: glacial acetic acid = 3:1) at 4℃ for 30 minutes. After being macerated in 1 mol/L HCl at 60 ℃ for three minutes, they were stained in 1% aceto-orcein and squashed.

Karyomorphological classification of interphase nuclei and mitotic prophase chromosomes followed Tanaka(1977,1971). The symbols for the description of karyotypes basically followed Levan *et al*.(1964): m = median-centromeric chromosome with arm ratio of 1.01 ~ 1.70; sm = submedian-centromeric chromosome with arm ratio of 1.71 ~ 3.00; st = subterminal-centromeric chromosome with arm ratio of 3.01 ~ 7.00; t = terminal-centromeric chromosome with arm ratio of over 7.01. The classification of karyotype asymmetry followed Stebbins (1971).

3 Results

3.1 Karyomorphology of interphase nuclei and mitotic prophase chromosomes

The three species studied were similar in karyomorphology of interphase nuclei. In the interphase nuclei (Fig. 1: A), numerous chromocenters were observed. The other regions were also stained well but unevenly. Thus, the interphase nuclei belonged to the complex chromocenter type.

The three species studied were also similar in karyomorphology of mitotic prophase chromosomes. In the prophase chromosomes (Fig. 1: B), hetero- and eu-chromatic segments were distinguishable but their boundaries were not distinct and the heterochromatic segments were distributed in the distal and interstitial as well as proximal regions. Thus, the prophase chromosomes belonged to the interstitial type

Table 1 Locality information and chromosome number of the three species studied of the genus *Oxygraphis* Bunge

Species	Locality	Voucher	2n
O. glacialis	Alpine meadow, alt. 3900 m, Deqen County, Yunnan Province.	Yang Qin-er and Kong Hong-zhi 397	16
O. tenuifolia	Alpine meadow, alt. 4100 m, Zhongdian County, Yunnan Province	Yang Qin-er and Kong Hong-zhi 342	16
O. delavayi	Alpine meadow, alt. 3900 m, Weixi County, Yunnan Province	Yang Qin-er and Kong Hong-zhi 446	16

3.2 Metaphase chromosomes3.2.1 *O. glacialis* (Fischer ex DC.) Bunge

The chromosomes were counted to be 2n = 16 (Fig.1:C; Fig.2:A). The first, second, fourth and fifth chromosome pairs were median-centromeric, the third pair submedian-centromeric, and the sixth, seventh and eighth pairs subterminal-centromeric. The karyotype was formulated as 2n = 8m + 2sm + 6st. The ratio of the length of the largest chromosome to the smallest one was 2.04, and the proportion of chromosomes with arm ratio > 2:1 was 0.50, the karyotype was therefore of 2B type according to the degree of asymmetry. The chromosomes in the complement ranged from 8.90 μm to 4.37 μm in length, and the total length of the complement (2n = 16) was 104.63 μm, with the average length of the chromosomes being 6.54 μm.

3.2.2 *O. tenuifolia* W. E. Evans

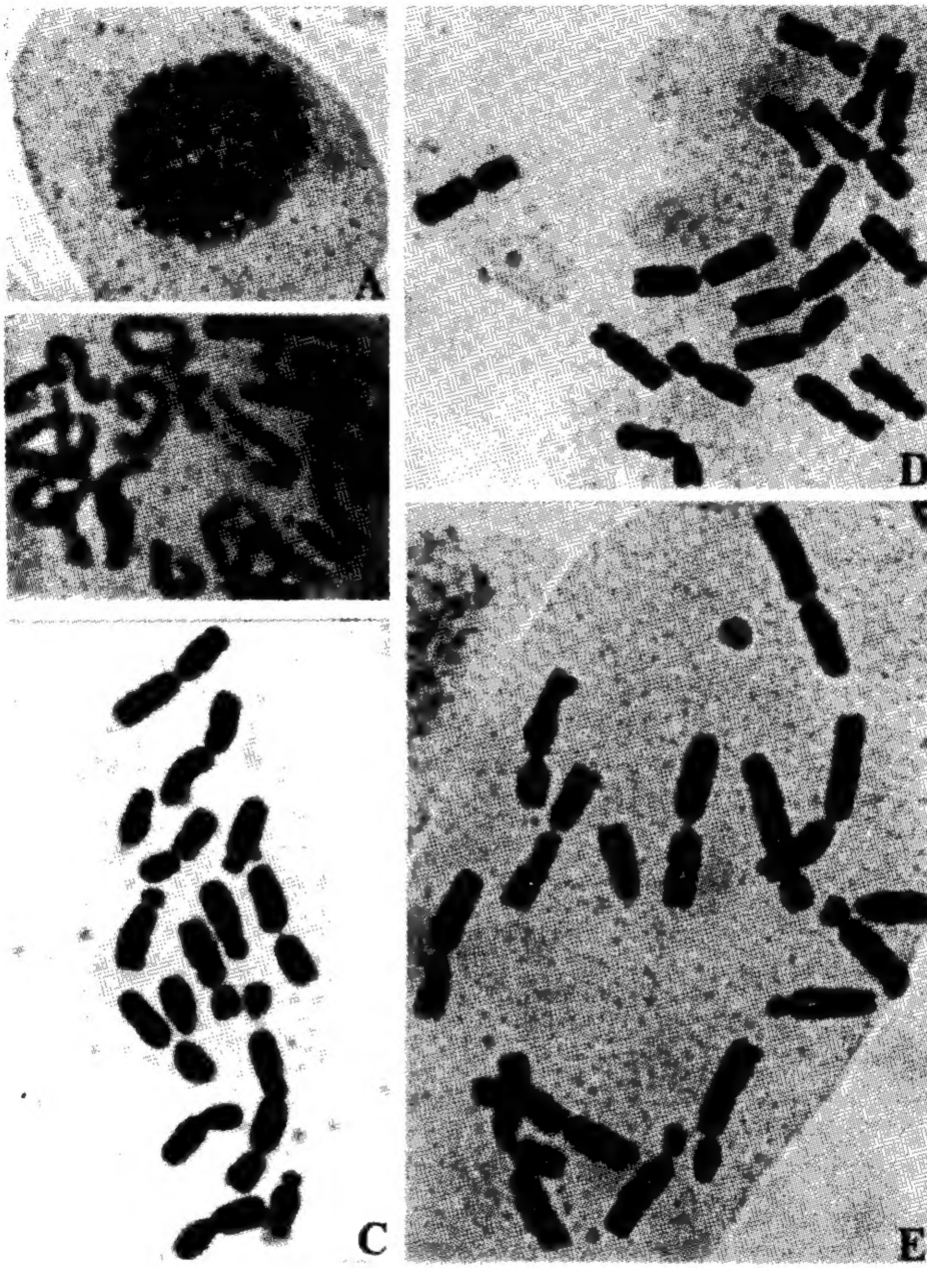

Fig.1 Chromosomes in three species of the genus *Oxygraphis* Bunge
A (interphase), B (prophase), C (metaphase): *O. glacialis*.
D (metapgase): *O. tenuifolia*. E (metaphase): *O. delavayi*. (all × 2030)

The chromosomes were counted to be 2n = 16 (Fig.1: D; Fig.2: B). The first, second and third chromosome pairs were median-centromeric, the fourth and fifth pairs submedian-centromeric, the sixth and seventh pairs subterminal-centromeric and the eighth pair terminal-centromeric. The karyotype was formulated as 2n = 6m + 4sm + 4st + 2t. The ratio of the length of the largest chromosome to the smallest one was 1.90, and the proportion of chromosomes with arm ratio > 2:1 was

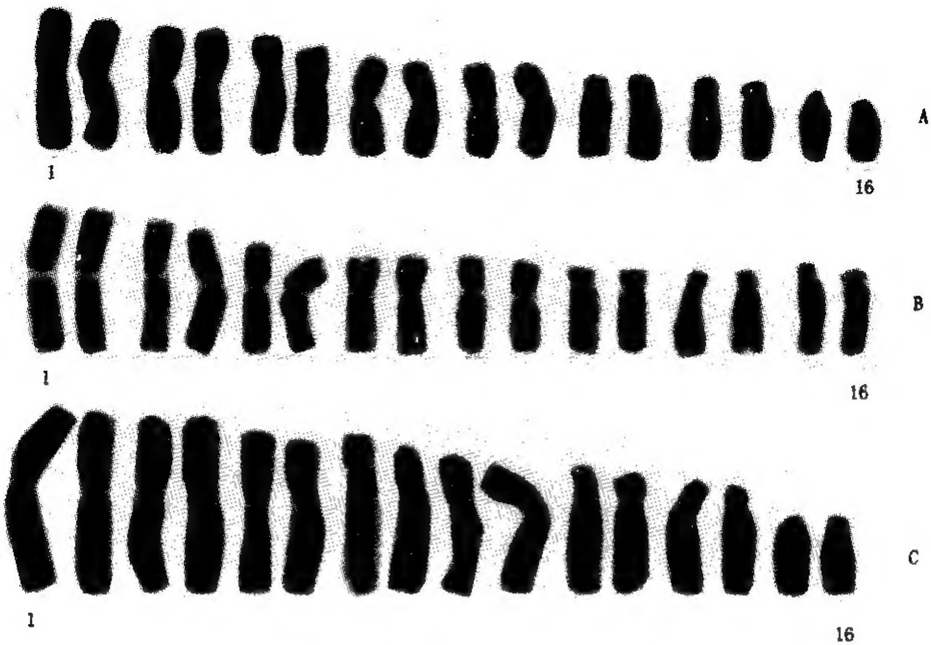

Fig.2 Karyotypes of three species of the genus *Oxygraphis* Bunge
A: *O. glacialis*. B: *O. tenuifolia*. C: *O. delavayi*. (all × 2030)

0.38, the karyotype was therefore of 2A type according to the degree of asymmetry. The chromosomes in the complement ranged from 9.36 μm to 4.93 μm in length, and the total length of the complement (2n = 16) was 103.37 μm, with the average length of the chromosomes being 6.46 μm. The chromosome number of this species is reported here for the first time.

3.2.3 *O. delavayi* Franch.

The chromosomes was counted to be 2n = 16 (Fig.1: E; Fig.2: C). The first, second and third chromosome pairs were median-centromeric, the fourth and fifth pairs submedian-centromeric, the sixth and seventh pairs subterminal-centromeric and the eighth pair terminal-centromeric. The karyotype was formulated as 2n = 6m + 4sm + 4st + 2t. The ratio of the length of the largest chromosome to the smallest one was 2.32, and the proportion of chromosomes with arm ratio > 2:1 was 0.50, the karyotype was therefore of 2B type according to the degree of asymmetry. The chromosomes in the complement ranged from 12.44 μm to 5.36 μm in length, and the total length of the complement (2n = 16) was 147.59 μm, with the average length of the chromosomes being 9.22 μm. The chromosome number of this species is reported here for the first time.

4 Discussion

4.1 The chromosomes of the three species studied in the genus *Oxygraphis* were all counted to be 2n = 16, thus confirming the previous reports that the basic chromosome number of this genus is x = 8 (Yang, Wu, 1993; Johnson, Parker, 1968).

4.2 As aforementioned, the karyotype of *O. glacialis* was previously reported consisting of 6 m, 6 sm and 4 st chromosomes, with the eighth chromosome pair having submedian centromeres (Yang, Wu, 1993). My result has shown that this chromosome pair had subterminal centromeres. Considering that the eighth chromosome pair of those species with x = 8 in the subtribe Ranunculinae is usu-

ally subterminal- or terminal-centromeric (Goepfert, 1974; Kurita, 1957), the result reported by Yang and Wu (1993) can be safely said to be wrong with respect to the morphology of the eighth chromosome pair.

4.3 Basically, the karyotypes of the three species studied were similar to each other in chromosome morphology, but that of *O. delavayi* was different in chromosome size. The chromosomes of this species were markedly larger than those of *O. glacialis* and of *O. tenuifolia*. In the Ranunculaceae, according to Kurita (1958), the karyotypes consisting of larger chromosomes might be more primitive than those of smaller ones. Gross-morphologically, *O. delavayi* is unique in the genus in having 3-lobed or occasionally 3-parted basal leaves and thinner deciduous sepals. Wang (1995) considered that these features might represent the primitive characters in the genus *Oxygraphis* and thus *O. delavayi* might be its primitive species. The chromosomal evidence supports this viewpoint.

4.4 In size and morphology, the chromosomes of the three species studied of *Oxygraphis* are basically similar to those of the species with $x = 8$ of the genera in the subtribe Ranunculinae (Goepfert, 1974; Kurita, 1957). This strongly supports the treatment of *Oxygraphis* as a member of this subtribe. Indeed, as stressed by Tamura (1995), karyological characters are of the utmost importance in the phylogenetic classification of the Ranunculaceae.

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摘要 研究了毛茛科鸭跖花属 *Oxygraphis* Bunge 3 种植物的核形态。其间期核均为复杂中央染色微粒型,前期染色体为中间型,染色体基数为 $x = 8$。脱萼鸭跖花 *O. delavayi* 的染色体明显大于鸭跖花 *O. glacialis* 和细叶鸭跖花 *O. tenuifolia* 的染色体。结果支持鸭跖花属是毛茛科毛茛亚族 subtrib. Ranunculinae 的一个自然成员以及脱萼鸭跖花可能是该属的原始种的观点。

关键词 染色体; 鸭跖花属

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